

AMENDMENTS

Listing of Claims

The following listing of claims replaces all previous listings or versions thereof:

1. (Previously presented) A method for determining the relative ratio of at least two different lipoproteins or apolipoproteins in a biological sample comprising:

immersing into the sample a solid phase material having separately immobilized thereon at least first and second antibody molecules, wherein the antibody molecules are selected from the group consisting of monoclonal antibodies, recombinant antibodies and antigen-binding antibody fragments thereof, wherein the antibody molecules are immunoreactive with at least two different lipoproteins, wherein the first and second antibodies bind to different stable, conformation independent epitopes that are uninfluenced by the lipid content of the lipoprotein, protein component of the lipoprotein or lipid associated with the specific lipoprotein, wherein the lipoproteins are selected from the group consisting of LDL, HDL and VLDL;

allowing the antibody molecules time to bind to the LDL, HDL, VLDL or apolipoproteins in the sample;

removing the solid phase material containing the immobilized antibody molecules;

determining the amount of lipoprotein or apolipoproteins bound by the immobilized antibody molecules, and

comparing the amount bound which is specific for LDL, HDL, VLDL or each apolipoprotein in order to calculate the relative amounts of LDL, HDL, VLDL or apolipoproteins.

2. (Previously presented) The method of claim 1 wherein the antibody molecules immobilized on the solid phase material are immunoreactive with lipoproteins selected from the group consisting of HDL and LDL.

3. (Previously presented) The method of claim 2 wherein the antibodies to the HDL or LDL are selected from the group consisting of recombinant antibodies and antibody fragments.

4. (Previously presented) The method of claim 3, wherein the first or second monoclonal antibodies are the anti-LDL monoclonal antibody produced by the hybridoma cell line HB₃cB₃ ATCC designation number HB 11612.

5. (Previously presented) The method of claim 3, wherein the first or second monoclonal antibodies are recombinant anti-LDL RcB₃M₁D₄ATCC designation number 69602.

6. (Three times amended) The method of claim 1 further comprising determining the amount of lipoprotein lipid or lipid associating with apolipoprotein by staining of the material bound to the immobilized antibody using a lipid stain.

7. (Original) The method of claim 6 wherein the lipid stain is selected from the group consisting of Sudan Red 7B, Oil Red O, and Sudan Black B.

8. (Original) The method of claim 6 wherein the lipoprotein lipid is stained prior to immersing the immobilized antibodies.

9. (Previously presented) The method of claim 6 further comprising measuring the amount of apolipoprotein or protein associated with the lipid in the sample, further comprising the step of providing antibodies immunoreactive with at least one apolipoprotein, wherein the antibodies are coupled to a protein stain, and staining the apolipoprotein or protein associated

with the lipid in the sample by reacting the protein stain coupled antibodies with the apolipoprotein or protein associated with the lipid in the sample.

10. (Original) The method of claim 1, wherein the apolipoprotein is selected from the group consisting of Apo A-I, Apo A-II, Apo B, Apo C-III, and Apo E.

11. (Original) The method of claim 1, wherein the biological sample is selected from the group consisting of blood, plasma, and serum.

12-38. (Canceled)

39. (Previously presented) A method for determining the relative ratio of LDL to HDL in a biological sample comprising

(a) determining the amount of LDL in the sample by

adding to the sample monoclonal antibody molecules immunoreactive with low density lipoprotein and not cross-reactive with high density lipoprotein and determining the amount of low density lipoprotein;

(b) determining the amount of HDL in the sample by

adding to the sample monoclonal antibody molecules immunoreactive with high density lipoprotein and not cross-reactive with low density lipoprotein and determining the amount of high density lipoprotein; and

(c) determining the ratio of the amount of low density lipoprotein with the amount of high density lipoprotein, wherein at least one of the monoclonal antibodies to LDL and HDL bind a stable, conformation independent epitope that is uninfluenced by the lipid content of the lipoprotein, the protein component of the lipoprotein or lipid associated with the specific lipoprotein.

40. (Canceled)

41. (Previously presented) A method for determining the relative ratio of VLDL to HDL comprising

(a) determining the amount of VLDL in the sample by
determining the amount of Apo E present in the VLDL in the sample by
providing Pan B antibody which is characterized by an equal binding and high affinity for all Apo B-containing lipoproteins in human plasma,

providing monoclonal antibody which specifically binds to Apo E associated with VLDL, contacting the antibodies reactive with Apo E associated with VLDL with the biological sample to form complexes between the anti-ApoE antibodies and Apo E containing particles,

contacting Pan B antibody with the biological sample containing the complexes between the anti-ApoE antibodies and ApoE containing particles to form complexes of anti-ApoB-anti-ApoE-ApoE containing particles, and

determining the amount of Apo E in the complexes of anti-ApoB-anti-ApoE-ApoE containing particles, which is the Apo E present in VLDL in the sample;

(b) removing the complexes of anti-ApoB-anti-ApoE-ApoE containing particles, either by binding of the anti-Apo E antibodies to an immobilized surface or centrifugation of sample to remove the complexes of anti-ApoB-anti-ApoE-ApoE containing particles;

and

(c) determining the amount of HDL in the sample by
determining the amount of Apo E present in the HDL in the sample by
providing Apo A-I monoclonal antibody immunoreactive specifically with Apo A-I,

contacting antibodies reactive with Apo E in HDL particles with the biological sample to form complexes between the anti-ApoE antibodies and Apo E containing particles,

contacting the Apo A-I monoclonal antibody with the biological sample to form complexes of the anti-ApoE antibodies-ApoE containing particles-anti-ApoA-I,

determining the amount of Apo E present in HDL in the complexes of the anti-ApoE antibodies-ApoE containing particles-anti-Apo A-I in the sample, and

determining the ratio of Apo E present in VLDL in the sample and Apo E present in HDL in the sample which is the ratio of VLDL to HDL,

wherein at least one of the monoclonal antibodies bind to a stable, conformation independent epitope that is uninfluenced by the lipid content of the lipoprotein, protein component of the lipoprotein or lipid associated with a specific lipoprotein selected from the group consisting of Apo B, Apo AI, and Apo E.

42. (Previously presented) A kit for determining the relative ratio of VLDL to HDL comprising

Pan B antibody which is characterized by an equal binding and high affinity for all Apo B-containing lipoproteins in human plasma,

monoclonal or recombinant antibody specifically immunoreactive with Apo C-III, and

monoclonal or recombinant Apo A-I antibody specifically immunoreactive with Apo A-I,

wherein at least one of the monoclonal or recombinant antibodies specifically bind to a stable, conformation independent epitope of a lipoprotein including Apo C-III or Apo A-I that is uninfluenced by the lipid content of the lipoprotein, protein component thereof or lipid

associated with a specific lipoprotein selected from the group consisting of Apo AI, and Apo CIII.

43. (Previously presented) The kit of claim 42 wherein the anti-Apo C-III or anti-A-I monoclonal or recombinant antibody molecules are selected from the group consisting of monoclonal antibodies, recombinant antibodies, and antigen binding antibody fragments thereof that specifically bind to a stable, conformation independent epitope which is uninfluenced by the lipid content of the lipoprotein, protein component thereof, or lipid associated with a specific lipoprotein.

44. (Previously presented) A kit for determining the relative ratio of VLDL to HDL comprising

Pan B antibody which is characterized by an equal binding and high affinity for all Apo B-containing lipoproteins in human plasma,

monoclonal antibody which binds to Apo E associated with VLDL ,

monoclonal Apo A-I antibody specifically immunoreactive with Apo A-I, and

monoclonal antibody which binds to Apo E in HDL,

wherein at least one of the antibodies binds to a stable, conformation independent epitope of a lipoprotein containing Apo E or Apo A-I that is uninfluenced by the lipid content of the lipoprotein, protein component of the lipoprotein or lipid associated with a specific lipoprotein.

45. (Previously presented) The kit of claim 44 wherein the anti-Apo E or anti-Apo A-I monoclonal antibody molecules are selected from the group consisting of monoclonal antibodies, recombinant antibodies, and monoclonal antibody fragments.

46. (Previously presented) A kit for determining the relative ratio of LPA-I and LPA-II lipoprotein particles comprising

monoclonal or recombinant Apo-A-I antibody specifically immunoreactive with Apo A-I lipoproteins in human plasma; and

monoclonal or recombinant Apo A-II antibody specifically immunoreactive with Apo A-II,

wherein the anti-Apo A-I or anti-Apo A-II monoclonal or recombinant antibody molecules are selected from the group consisting of monoclonal antibodies, recombinant antibodies, and antigen-binding antibody fragments thereof that specifically bind to a stable, conformation independent epitope of a lipoprotein containing Apo A-I or Apo A-II which is uninfluenced by the lipid content of the lipoprotein, protein component of the lipoprotein, or lipid associated with a specific lipoprotein.

47. (Previously presented) The kit of claim 46 wherein the anti-Apo A-I and anti-Apo A-II monoclonal or recombinant antibody molecules are selected from the group consisting of monoclonal antibodies, recombinant antibodies, and monoclonal antibody fragments that specifically bind to a stable, conformation independent epitope which is uninfluenced by the lipid content of the lipoprotein, protein component of the lipoprotein, or lipid associated with a specific lipoprotein.